CLAIMS

We claim:

1. A culture of *Sphingomonas* sp. strain AD109 or a mutant thereof.

2. A nucleotide molecule encoding an enzyme having an amino acid sequence set forth in SEQ ID NO.: 2; or a mutant, fragment or homologue thereof.

3. The nucleotide molecule of Claim 2 having substantially the same sequence as the sequence set forth in SEQ ID NO.: 1.

4. A nucleotide molecule encoding an enzyme having an amino acid sequence set forth in SEQ ID NO.: 4; or a mutant, fragment or homologue thereof.

5. The nucleotide molecule of Claim 4 having substantially the same sequence set forth in SEQ ID NO.: 3.

6. A nucleotide molecule encoding an enzyme having an amino acid sequence set forth in SEQ ID NO.: 6; or a mutant, fragment or homologue thereof.

7. The nucleotide molecule of Claim 6 having substantially the same sequence as the sequence set forth in SEQ ID NO.: 5.

8. A nucleic acid molecule comprising the nucleotide sequence set forth in SEQ ID NO.: 1, SEQ ID NO.: 3, or SEQ ID NO.: 5; a mutant or fragment thereof; or a combination thereof.

8. A nucleotide sequence comprising at least about 20 contiguous nucleotides from the sequence of SEQ ID NO.: 1, or the complement thereof.

9. The nucleotide sequence of Claim 8 comprising at least about 40 contiguous nucleotides from the sequence of SEQ ID NO.: 1 or the complement thereof.

10. The nucleotide sequence of Claim 8 comprising at least about 50 contiguous nucleotides from the sequence of SEQ ID NO.: 1 or the complement thereof.

11. A nucleotide sequence comprising at least about 20 contiguous nucleotides from the sequence of SEQ ID NO.: 3, or the complement thereof.

12. The nucleotide sequence of Claim 11 comprising at least about 40 contiguous nucleotides from the sequence of SEQ ID NO.: 3 or the complement thereof.

13. The nucleotide sequence of Claim 11 comprising at least about 50 contiguous nucleotides from the sequence of SEQ ID NO.: 3 or the complement thereof.

14. A nucleotide sequence comprising at least about 20 contiguous nucleotides from the sequence of SEQ ID NO.: 5, or the complement thereof.

15. The nucleotide sequence of Claim 14 comprising at least about 40 contiguous nucleotides.

16 ~~45~~ 70 14

~~17~~. The nucleotide sequence of Claim ~~15~~ comprising at least about 50 contiguous nucleotides.

~~18~~. A nucleotide sequence which specifically hybridizes to a polynucleotide molecule comprising the nucleotide sequence set forth in SEQ ID NO.: 1.

19. The nucleotide sequence of Claim 18, wherein the isolated nucleotide sequence hybridizes to the polynucleotide molecule under conditions of high stringency.

~~20~~. A nucleotide sequence which hybridizes to a polynucleotide molecule comprising the nucleotide sequence set forth in SEQ ID NO.: 3.

21. The nucleotide sequence of Claim 20, wherein the isolated nucleotide sequence hybridizes to the polynucleotide molecule under conditions of high stringency.

~~22~~. A nucleotide sequence which hybridizes to a polynucleotide molecule comprising the nucleotide sequence set forth in SEQ ID NO.: 5.

23. The nucleotide sequence of Claim 22, wherein the isolated nucleotide sequence hybridizes to the polynucleotide molecule under conditions of high stringency.

~~24~~. An enzyme having substantially the amino acid sequence set forth in SEQ ID NO.: 2, or an enzymatically active fragment thereof.

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~~25~~. The enzyme of Claim ~~24~~, wherein said enzyme is isolated from a microorganism.

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~~26~~. The enzyme of Claim ~~25~~ wherein the microorganism is a *Sphingomonas*.

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~~27~~. The enzyme of Claim ~~26~~ wherein the microorganism is *Sphingomonas* sp. strain AD109.

28. The enzyme of Claim 27 having substantially the amino acid sequence set forth in SEQ ID NO.: 2, or fragment thereof, said enzyme being substantially free of other *Sphingomonas* proteins.

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~~29~~. An enzyme having substantially the amino acid sequence set forth in SEQ ID NO.: 4, or an enzymatically active fragment thereof.

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~~30~~. The enzyme of Claim ~~29~~, wherein said enzyme is isolated from a microorganism.

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~~31~~. The enzyme of Claim ~~30~~ wherein the microorganism is a *Sphingomonas*.

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~~32~~. The enzyme of Claim ~~31~~ wherein the microorganism is *Sphingomonas* sp. strain AD109.

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~~33~~. The enzyme of Claim ~~32~~ having a molecular weight of about 40,000 daltons.

~~34~~. An enzyme having substantially the amino acid sequence set forth in SEQ ID NO.: 4, or fragment thereof, said enzyme being substantially free of other *Sphingomonas* proteins.

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~~35~~. An enzyme having substantially the amino acid sequence set forth in SEQ ID NO.: 6, or an enzymatically active fragment thereof.

29 ~~36~~. The enzyme of Claim ~~35~~ 28, wherein said enzyme is isolated from a microorganism.

30 ~~37~~. The enzyme of Claim ~~36~~ 29 wherein the microorganism is a *Sphingomonas*.

31 ~~38~~. The enzyme of Claim ~~37~~ 30 wherein the microorganism is *Sphingomonas* sp. strain AD109.

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~~39~~. An enzyme having substantially the amino acid sequence set forth in SEQ ID NO.: 6, or fragment thereof, said enzyme being substantially free of other *Sphingomonas* proteins.

33 ~~40~~. An enzyme comprising the amino acid sequence set forth in SEQ ID NO.: 2.

34 ~~41~~. An enzyme comprising the amino acid sequence set forth in SEQ ID NO.: 4.

35 ~~42~~. An enzyme comprising the amino acid sequence set forth in SEQ ID NO.: 6.

36 ~~43~~. A *Sphingomonas* enzyme catalyzing the conversion of dibenzothiophene to dibenzothiophene-5,5-dioxide.

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~~44~~. A *Sphingomonas* enzyme catalyzing the conversion of dibenzothiophene-5,5-dioxide to 2-(2-hydroxyphenyl)benzenesulfinate.

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~~45~~. A *Sphingomonas* enzyme catalyzing the conversion of 2-(2-hydroxyphenyl)benzenesulfinate to 2-hydroxybiphenyl and inorganic sulfur.

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~~46~~. A plasmid comprising a nucleic acid molecule of Claim ~~2~~ 1 operatively linked to a promoter.

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~~47~~. A plasmid comprising a nucleic acid molecule of Claim ~~4~~ 3 operatively linked to a promoter.

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~~48~~. A plasmid comprising a nucleic acid molecule of Claim ~~6~~ 5 operatively linked to a promoter.

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~~49~~. A plasmid comprising a nucleotide molecule of Claim ~~8~~ 7 operatively linked to a promoter.

50. A transformed microorganism containing a recombinant DNA plasmid comprising a DNA molecule encoding an enzyme set forth in SEQ ID NO.: 2, a mutant or a homologue thereof.

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51. A transformed microorganism containing a recombinant DNA plasmid comprising a DNA molecule encoding an enzyme having the sequence set forth in SEQ ID NO.: 4, or a mutant, fragment or homologue thereof.

52. A transformed microorganism containing a recombinant DNA plasmid comprising a DNA molecule encoding an enzyme having the amino acid sequence set forth in SEQ ID NO.: 6; or a mutant or homologue thereof.

53. A transformed microorganism containing a recombinant DNA plasmid comprising a DNA sequence encoding an enzyme having the sequence set forth in SEQ ID NO.: 2, or a mutant, fragment or homologue thereof; a DNA sequence encoding an enzyme having the sequence set forth in SEQ ID NO.: 4, or a mutant, fragment or homologue thereof; and a DNA sequence encoding an enzyme having the amino acid sequence set forth in SEQ ID NO.: 6, or a mutant, fragment or homologue thereof.

54. A method of desulfurizing a fossil fuel containing organosulfur molecules, comprising the steps of:

(a) contacting the fossil fuel with an aqueous phase containing a *Sphingomonas*-derived desulfurization biocatalyst thereby forming a fossil fuel and aqueous phase mixture;

(b) maintaining the mixture under conditions sufficient for desulfurization, thereby resulting in a fossil fuel having a reduced organic sulfur content; and

(c) separating the fossil fuel having a reduced organic sulfur content from the resulting aqueous phase.

55. The method of Claim 54 wherein the biocatalyst comprises an enzyme preparation or a microorganism comprising an enzyme having the sequence set forth

in SEQ ID NO.: 2, or a mutant, active fragment or homologue thereof; an enzyme having the sequence set forth in SEQ ID NO.: 4, or a mutant, active fragment or homologue thereof; and an enzyme having the amino acid sequence set forth in SEQ ID NO.: 6, or a mutant, active fragment or homologue thereof.

56. The method of Claim 55 further comprising the steps of adding a flavoprotein, flavin, NADH or a combination thereof.

57. The method of Claim 55 wherein the fossil fuel is a liquid hydrocarbon.

58. The method of Claim 57 wherein the liquid hydrocarbon ~~is~~ a petroleum.

59. The method of Claim 55 wherein the biocatalyst is a microorganism.

60. The method of Claim 59 wherein the microorganism is *Sphingomonas* sp. strain AD109.

61. The method of Claim 59 wherein the microorganism contains a heterologous DNA molecule which encodes the biocatalyst.

62. The method of Claim 61 wherein the heterologous DNA is derived from *Sphingomonas* sp. strain AD109.

63. The method of Claim 55 wherein the biocatalyst is a cell-free fraction.

64. The method of Claim 63 wherein the biocatalyst is a cell-free fraction of *Sphingomonas* sp. strain AD109.

65. A method of oxidizing organic molecules, comprising the steps of:

(a) contacting the organic molecules with an aqueous phase containing a *Sphingomonas*-derived biocatalyst capable of oxidizing organosulfur compounds, thereby forming an organic compound and aqueous phase mixture; and

(b) maintaining the mixture under conditions sufficient for oxidation of the organic molecules by the biocatalyst, thereby forming an oxidized organic compound.

66. The method of Claim 65 wherein the biocatalyst comprises an enzyme having the sequence set forth in SEQ ID NO.: 2, or a mutant, active fragment or homologue thereof; an enzyme having the sequence set forth in SEQ ID NO.: 4, or a mutant, active fragment or homologue thereof; an enzyme having the amino acid sequence set forth in SEQ ID NO.: 6, or a mutant, active fragment or homologue thereof; or a combination thereof.

67. The method of Claim 66 wherein the organic is an organosulfur compound which is a component of a fossil fuel.

68. The method of Claim 67 wherein the organosulfur compound is a substituted or unsubstituted dibenzothiophene and the oxidized organosulfur compound is a substituted or unsubstituted dibenzothiophene-5-5-dioxide.

69. The method of Claim 67 wherein the organosulfur compound is a substituted or unsubstituted dibenzothiophene-5-5-dioxide and the oxidized organosulfur compound is a substituted or unsubstituted 2-(2-hydroxyphenyl)benzenesulfinate.

70. The method of Claim 66 wherein the biocatalyst capable of oxidizing organosulfur molecules is a microorganism.

71. The method of Claim 70 wherein the microorganism is *Sphingomonas* sp. strain AD109.

72. The method of Claim 70 wherein the microorganism contains a heterologous DNA molecule which encodes the biocatalyst.

73. The method of Claim 72 wherein the heterologous DNA is derived from *Sphingomonas* sp. strain AD109.

74. The method of Claim 66 wherein the biocatalyst capable of oxidizing organosulfur compounds is a cell-free fraction.

75. The method of Claim 74 wherein the biocatalyst is a cell-free fraction of *Sphingomonas* sp. strain AD109.

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